THE INFLUENCE OF SELENIUM AND SALINITY SOURCES ON SELENIUM UPTAKE BY ALFALFA: A GREENHOUSE SAND CULTURE POT EXPERIMENT

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Personnel
P. F. Pratt
N. Albasel
H. Wan

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SUMMARY

Alfalfa (*Medicago sativa* L.) was grown in greenhouse sand culture to examine the effect of Se, salinity and salinity composition on Se accumulation by plants. In a 2x2x4 factorial experiment, salinity was added as either SO_4^{2-} or Cl^- salts in the irrigating solution to achieve electrical conductivity of 0.50, 0.75, 1.00, and 1.25. Selenium was added to the nutrient solution at concentrations of 125 or 250 ug Se(VI) L^{-1}. After the third harvest, the roots were washed and all plant materials were analyzed for dry weight and Se content. Plant yields were not significantly affected by either low rates of salinity or Se applications. Plant Se accumulation was reduced from 42 to 3 mg kg^{-1}, from 161 to 4 mg kg^{-1}, and from 160 to 4 mg kg^{-1} in the first, second and third cuttings, respectively, in the presence of SO_4^{2-} salinity (0.5 mmol L^{-1} to 1.25 mmol L^{-1}) due to an apparent SO_4^{2-}-Se(VI) antagonism. Salinity source as Cl^- was less effective than SO_4^{2-}. In the west side of the San Joaquin Valley (SJV), however, soils and waters that contain high levels of Se also contain high levels of SO_4^{2-}. The presence of SO_4^{2-} with Se reduces the plant accumulation of Se and thus lowers the possibility of potential hazard of animals feeding on crops growing in areas enriched with Se.

INTRODUCTION

In California, Se-rich soils are found at lower elevations on the west side of the San Joaquin Valley. While a number of conditions may lead to the production of seleniferous soils, not all Se-rich soils are associated with diseases of Se toxicity (Ganje, 1966). The key to the problem of soil toxicity is the chemical state of the Se which is present. Selenate Se(VI) is the predominant form of Se in the liquid phase of alkaline soils. The solubility of Se in most soils is rather low; therefore, many agricultural areas produce crop plants and forages with low Se contents.

The water-soluble fraction of soil Se is considered to be the fraction that is available to plants. Van Dorst and Peterson (1982) reported that the behavior of Se in highly calcareous soil is of special concern because when soils are low in sesquioxides the Se becomes easily water-soluble. Singh (1982) stated that the best effects in correcting Se toxicity to plants in such soils are obtained by the application of S, P and even N on the soils. Selenium is essential for animal nutrition in trace amounts, but can become potentially hazardous for livestock when present in forage crops at higher concentrations. Interest in the accumulation of Se by plants has been stimulated by the discovery of Se concentrations as high as 3800 ug Se L^{-1} in agricultural drainage water in California (Deverel et al., 1984). In addition to Se, this drainage water also contains a mixture of Cl^- and SO_4^{2-} salts. The salinity in these waters based on electrical conductivity measurements commonly ranged from 431 to 68,000 umho/cm, and with the median of 3,655 umho/cm. The sulfate concentrations ranged from 39 to 65,000 mg/L and with the median value of 1,700 mg/L. The chloride concentration ranged from 29 to 16,000 mg/L and with the median value of 290 mg/L (Deverel et al., 1984).

The uptake and accumulation of Se by plants is influenced by many factors including the presence of other ions in the soil solution (Mikkelsen et al., 1987). The close relationship existing between Se(VI) and SO_4^{2-} for plant uptake has been observed by several workers (Gupta and Winter, 1975; Gissel-Nielsen et al., 1984). The reuse of this high Se drainage water for irrigation by using salt tolerant crop species, blending saline water with low-salt water, and adequate irrigation management, raises several questions regarding the potential accumulation of Se by irrigated crops, particularly growing in the west side of San Joaquin Valley.

The main objectives of the present study were (1) to determine the effect of Se and two sources of salinity (SO_4^{2-} and Cl^-) on the Se uptake by alfalfa plants, (2) to determine whether the Se conclusions could be confirmed in the range of irrigation water being used in the west side of the SJV, and (3) to help establish a Se water quality guideline for the protection of crops and livestock under the specific conditions in the west side of the San Joaquin Valley.

MATERIALS AND METHODS

Alfalfa (<u>Medicago sativa</u> L., Germain WL 512) was planted in a glasshouse into 11-L plastic pots filled with coarse-grained silica sand. Two pots were placed above one 110-L plastic-lined reservoir which contained nutrient solution. The plants were irrigated automatically five times each day by pumping the nutrient solution from the reservoir onto the surface of the pot and allowing it to slowly drain into the reservoir again. After emergence, each pot was thinned to 10 plants.

The basal nutrient solution for each reservoir contained the following: 0.5 mM KH_2PO_4, 1.25 mM $Ca(NO_3)_2$, 1.2 mM KNO_3, 0.2 mM $Mg(NO_3)_2$, 0.5 mM $MgSO_4 \cdot 7H_2O$, 5.0 mg Fe L^{-1} (EDDHA), 0.5 mg B L^{-1}, 0.5 mg Mn L^{-1}, 0.05 mg Zn L^{-1}, 0.02 mg Cu L^{-1}, and 0.01 mg Mo L^{-1}. The nutrient solutions were replenished midway through the experiment. The solution pH was maintained between pH 6.5 and 7.0 with additions of HCl or NaOH as needed. The electrical conductivity of the base nutrient solution was 0.5 dS m^{-1}.

A factorial experiment (2x2x4) was imposed in three replications in a completely randomized block design 10 days after plant emergence. The treatments consisted of 2 Se concentrations (125 and 250 ug L^{-1} as Na_2SeO_4), 2 salinity sources (Cl^- as one level of 0.125 dS m^{-1} equivalent amounts of $CaCl_2$ and NaCl; and 4 levels of sulphate (0.5, 0.75, 1.00, 1.25 dS m^{-1} and as mg SO_4^{2-} L^{-1}, these values are 209, 313, 417 and 522 mg SO_4^{2-} L^{-1}, respectively) as equivalent amounts of $CaSO_4 \cdot 2H_2O$, $MgSO_4$, and Na_2SO_4). A control with no added Se or salinity was also included. The treatment concentrations were monitored throughout the experiment and adjusted as needed to maintain the desired levels of salinity and Se.

Three cuttings of alfalfa were taken at the 0.25 bloom stage at 3 cm above the sand level. At the conclusion of the experiment, the roots were washed and separated from the sand. Plant yields were measured after drying at 50°C prior to grinding and chemical analysis. Plant Se was determined from tissue digested with nitric and perchloric acids (Ganje and Page, 1974) and analyzed with atomic absorption hydride generation.

RESULTS AND DISCUSSION

Yield

The effect of both factors in the treatment solutions, i.e., salinity and Se, on the plant yield were not significantly pronounced (Table 1). However, slight differences were found between the control and the SO_4^{2-} and Se(VI) treatments and mainly for the first harvest. It seems that the reason for this lack of response in yield is probably the low rates of both factors, SO_4^{2-} and Se(VI) applied in the treatment solutions. This confirms the salinity tolerance data presented by Maas (1986). Soltanpour and Workman (1980) found that a remarkable yield reduction in alfalfa due to Se toxicity occurred when plant Se content exceeded 25-30 mg kg^{-1}. The Se contents of the SO_4^{2-} and Cl^- treated plants were consistently below this toxic level and thus decreases in yields were not expected.

Plant Selenium Content

Plant Se contents were affected by both the level and form of salinity as well as the concentration of Se in the treatment solution (Table 2). At the low level of Se (125 ug L^{-1}) additions, plant Se concentrations were reduced from 83 to 2 mg Se kg^{-1} in the presence of 0 to 1.75 dS m^{-1} SO_4^{2-} salinity (Table 2, Figures 1-3). At the higher rate of Se in the nutrient solution (250 ug Se kg^{-1}), SO_4^{2-} also had a dramatic effect on Se accumulation by alfalfa (Figures 1-3). This similar pattern of response shown at the lower rate of Se application supports the effect of SO_4^{2-} on reduction in uptake of Se by the plant (Table 2). This reduction was significantly pronounced in the first, second and the third cuttings (Table 3).

Similarly, Mikkelsen et al. (1987) reported that addition of 2,500 mg SO_4^{2-} L^{-1} (or 6 dS m^{-1}) to sand culture treating solutions with 1 mg Se(VI) L^{-1}, plant Se concentrations were reduced from over 600 mg Se kg^{-1} to less than 7 mg Se kg^{-1}. In the present experiment, the Se concentration of the plants increased with the level of Se in the irrigating solution and decreased with additional SO_4^{2-}. A lesser antagonistic interaction between Se and Cl^- salinity was also manifested in the Se concentrations in forage and in the root tissue (Tables 2 & 4 and Figures 1-4).

Due to the similarity between Se(VI) and SO_4^{2-}, it is not surprising that SO_4^{2-} depresses Se(VI) accumulation by plants. The antagonistic interaction between Se(VI) and SO_4^{2-} was first reported by Hurd-Karrer (1938) and later by Carter et al. (1969), Gupta and Winter (1975), and Partley and McFarlane. (1974). The nature of the Se(VI)-SO_4^{2-} antagonism is not completely understood but short-term experiments by Epstein (1955) and Ferrari and Renosto (1972) with excised barley roots showed that the uptake mechanism for these two anions was identical. They also reported that the affinity for uptake of these two anions is approximately equal. Therefore, as demonstrated in this experiment, plant-Se content decreases as the SO_4^{2-} to Se(VI) ratio increases, and as SO_4^{2-} becomes the dominant ion at the root uptake site at higher SO_4^{2-} x Se ratios.

The antagonistic interactions between Se(VI) and SO_4^{2-} for plant uptake presented in this experiment have current implications for irrigated agriculture. In many areas where Se concentrations are elevated in soils and waters, SO_4^{2-} is also abundant (Allaway, 1970). Thus, this approach strongly supports the new Se guidelines recently suggested (100 ug L^{-1}) (Pratt et al., 1988) for the water used for irrigation in the west side of the San Joaquin Valley. A survey of wells in California found that waters exceeding 250 ug Se(VI) L^{-1}, and containing over 700 mg SO_4 L^{-1}, were used for irrigation (Oster, 1987). The presence of abundant SO_4^{2-} has likely reduced the accumulation of Se by crops grown. Grattan et al. (1987) reported that crops accumulate Se when irrigated with drainage waters containing Se at 30 to 40 ug L^{-1} and 200 to 350 ug L^{-1} at West Side Field Station, and at Murrieta Farms in Mendota, respectively. The Se uptake, however, produced relatively low (6 to 8 ug Se) increases in human diet and was not considered a health hazard by Fan et al. (1987). Therefore, the antagonistic interactions between SO_4^{2-} x Se, as exhibited in this experiment and reported by other researchers, appears to play a major role in reducing Se uptake by plants.

The roots contain higher Se concentrations than the upper plant parts. This pattern of Se-plant distribution was in agreement with the data reported from field experiments by Grattan et al. (1987), Ayars and Letey (1987) and from similar sand culture pot experiment by Mikkelsen et al. (1987).

Conclusions

The uptake and accumulation of Se by plants is influenced by many factors including the presence of other ions. The close relationship between Se(VI) and SO_4^{2-} for plant uptake has been observed by others (Mikkelsen et al., 1987) as well as in the present study. Plant Se concentration was reduced from 42 to 3 mg Se kg^{-1}, from 161 to 4 mg Se kg^{-1}, and from 160 to 4 mg Se kg^{-1} in the first, second and third cuttings, respectively, in the presence of SO_4^{2-} salinity of 0.5 to 1.25 mmhos L^{-1} as added treatments, and these levels as total salinity as indicated in Tables 1-2 and Figures 1-4 are 0.5 to 1.75 dS m^{-1}.

As Part I of this report (under the Se section), a guideline of 100 ug Se L^{-1} was suggested for irrigation waters used on the west side of the San Joaquin Valley where waters also contain SO_4^{2-} ions. This study presented here support this recommendation, but also indicate that the previously recommended maximum Se concentration of 20 ug L^{-1} is probably appropriate for nonsaline waters or saline waters that contain very low SO_4^{2-} concentrations.

Table I. EFFECT OF Se AND SALINITY SOURCE (SO_4^{2-} AND Cl^-) AND LEVEL ON THE YIELD OF ALFALFA.

SELENIUM µg L^{-1}	SOURCE OF SALINITY	TOTAL SALINITY dS m^{-1}	YIELD (dry wt. basis) HARVEST NUMBER 1	2	3	ROOTS
			g/pot			
0	NUTRIENT SOLUTION (NS)	0.50	61.46	45.46	53.26	36.57
125	NS	0.50	41.85	43.22	44.79	26.54
	NS + SO_4	1.00	40.58	35.36	44.18	15.81
	NS + SO_4	1.25	47.77	44.86	62.76	27.53
	NS + SO_4	1.50	42.72	44.11	48.63	24.62
	NS + SO_4	1.75	55.37	43.32	48.55	29.73
$LSD_{0.05}$			16.531	13.657	8.254	11.765
250	NS	0.50	44.85	42.99	47.45	23.08
	NS + SO_4	1.00	63.90	49.14	58.54	25.18
	NS + SO_4	1.25	49.23	45.67	56.93	24.00
	NS + SO_4	1.50	40.93	36.89	41.49	23.42
	NS + SO_4	1.75	53.00	35.35	45.84	23.96
$LSD_{0.05}$			28.117	11.046	19.322	15.254
125	NS	0.50	60.46	41.11	43.98	29.92
	NS+SO_4+Cl‡	0.88	60.97	37.91	39.94	33.55
	NS+SO_4+Cl	1.12	61.55	46.71	49.76	30.73
	NS+SO_4+Cl	1.38	48.94	45.04	48.49	27.41
$LSD_{0.05}$			37.765	14.062	13.411	27.083

‡ CHLORIDE ADDITIONS INCREASED THE EC BY 0.125 dS m^{-1}. OTHER INCREASES WERE FROM SO_4 ADDITIONS.

Table 2. THE EFFECT OF Se AND SALINITY SOURCE AND LEVEL ON Se ACCUMULATION BY ALFALFA.

SELENIUM $\mu g\ L^{-1}$	SOURCE OF SALINITY	TOTAL SALINITY $dS\ m^{-1}$	Se (dry wt. basis) HARVEST NUMBER 1	2	3	ROOTS
			----------- mg/kg -----------			
0	NUTRIENT SOLUTION (NS)	0.50	0.04	0.07	0.08	0.08
125	NS	0.50	20.98	81.93	83.42	205.92
	NS + SO_4	1.00	3.53	4.55	4.73	14.72
	NS + SO_4	1.25	2.73	3.78	4.34	8.21
	NS + SO_4	1.50	2.39	3.02	3.20	4.31
	NS + SO_4	1.75	1.50	2.32	2.35	3.76
$LSD_{0.05}$			1.403	1.958	1.480	2.327
250	NS	0.50	41.61	161.41	159.62	360.4
	NS + SO_4	1.00	6.20	9.75	9.65	19.45
	NS + SO_4	1.25	4.53	6.35	7.08	15.0
	NS + SO_4	1.50	3.45	4.81	5.32	9.32
	NS + SO_4	1.75	2.84	4.07	4.43	7.11
$LSD_{0.05}$			1.655	1.830	1.310	2.930
125	NS	0.50	20.93	81.93	83.42	205.92
	NS + SO_4 + Cl‡	0.88	4.84	8.50	7.18	15.12
	NS + SO_4 + Cl	1.12	3.19	5.82	5.81	10.22
	NS + SO_4 + Cl	1.38	2.59	4.46	3.92	7.10
$LSD_{0.05}$			1.696	2.189	1.789	1.983

‡ CHLORIDE ADDITIONS INCREASED THE EC BY 0.125 $dS\ m^{-1}$. OTHER INCREASES WERE FROM SO_4 ADDITIONS.

Table 3. ANALYSIS OF VARIANCE SUMMARY OF THE EFFECTS OF SALINITY SOURCE (SO_4^{2-}) AND LEVEL, Se CONCENTRATION, AND THEIR INTERACTIONS ON THE Se CONCENTRATION OF ALFAFA.

HARVEST	SOURCE OF VARIATION	DEGREES OF FREEDOM	SS†	F RATIO	SIGNIFICANCE
	SO_4	4	3759	1132	*
1st	Se	1	227	273	*
	SO_4 x Se	4	431	130	*
	SO_4	4	65674	4105	*
2nd	Se	1	2450	1899	*
	SO_4 x Se	4	7080	443	*
	SO_4	4	65062	79344	*
3rd	Se	1	2325	11341	*
	SO_4 x Se	4	6449	7865	*
	SO_4	4	358187	19093	*
ROOTS	Se	1	9093	1939	*
	SO_4 x Se	4	26873	1317	*

* INDICATES 0.005 LEVEL OF PROBABILITY.

†SS, SUM OF SQUARES; F, VARIANCE RATIO.

Table 4. ANALYSIS OF VARIANCE SUMMARY OF THE EFFECTS OF SALINITY SOURCE (SO_4^{2-} & Cl^-) AND LEVEL, Se CONCENTRATION, AND THEIR INTERACTIONS ON THE Se CONCENTRATION OF ALFALFA.

HARVEST	SOURCE OF VARIATION	DEGREES OF FREEDOM	SS†	F RATIO	SIGNIFICANCE
1st	SO_4	2	1293	3233	**
	Cl	1	0.114	0.57	
	SO_4 x Cl	2	0.086	0.22	
2nd	SO_4	2	23915	3737	**
	Cl	1	2.24	0.7	
	SO_4 x Cl	2	1.26	0.2	
3rd	SO_4	2	24786	15991	**
	Cl	1	0.2	0.26	
	SO_4 x Cl	2	1.8	1.16	
ROOTS	SO_4	2	153505	30578	**
	Cl	1	15.71	6.26	*
	SO_4 x Cl	2	16.59	3.31	

*, ** INDICATES 0.05 AND 0.005 LEVEL OF PROBABILITY, RESPECTIVELY.

†SS, SUM OF SQUARE ; F, VARIANCE RATIO.

Fig. 1. EFFECTS OF SO_4 AND Cl ON Se UPTAKE BY ALFALFA

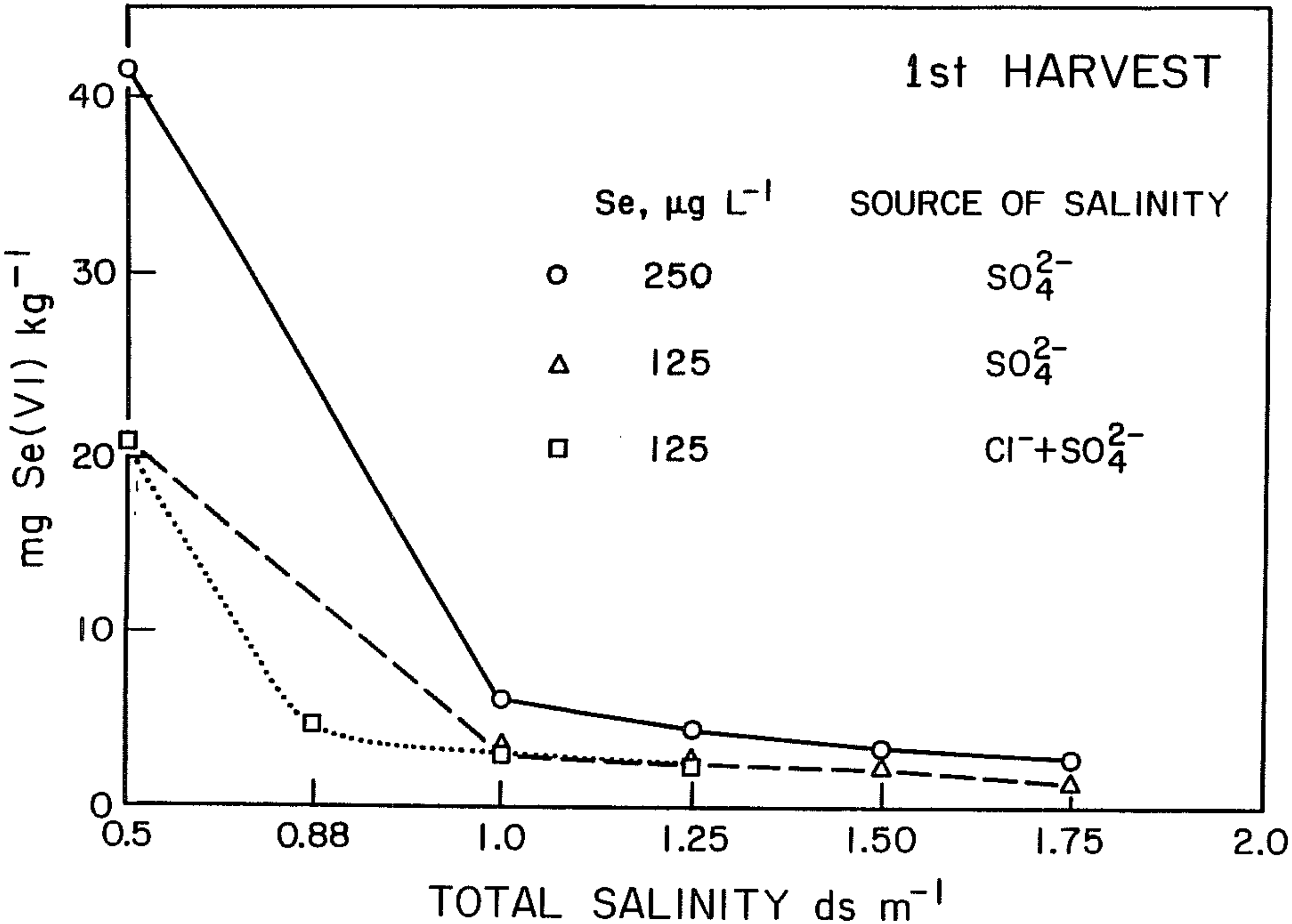


Fig. 2. EFFECTS OF SO_4 AND Cl ON Se UPTAKE BY ALFALFA

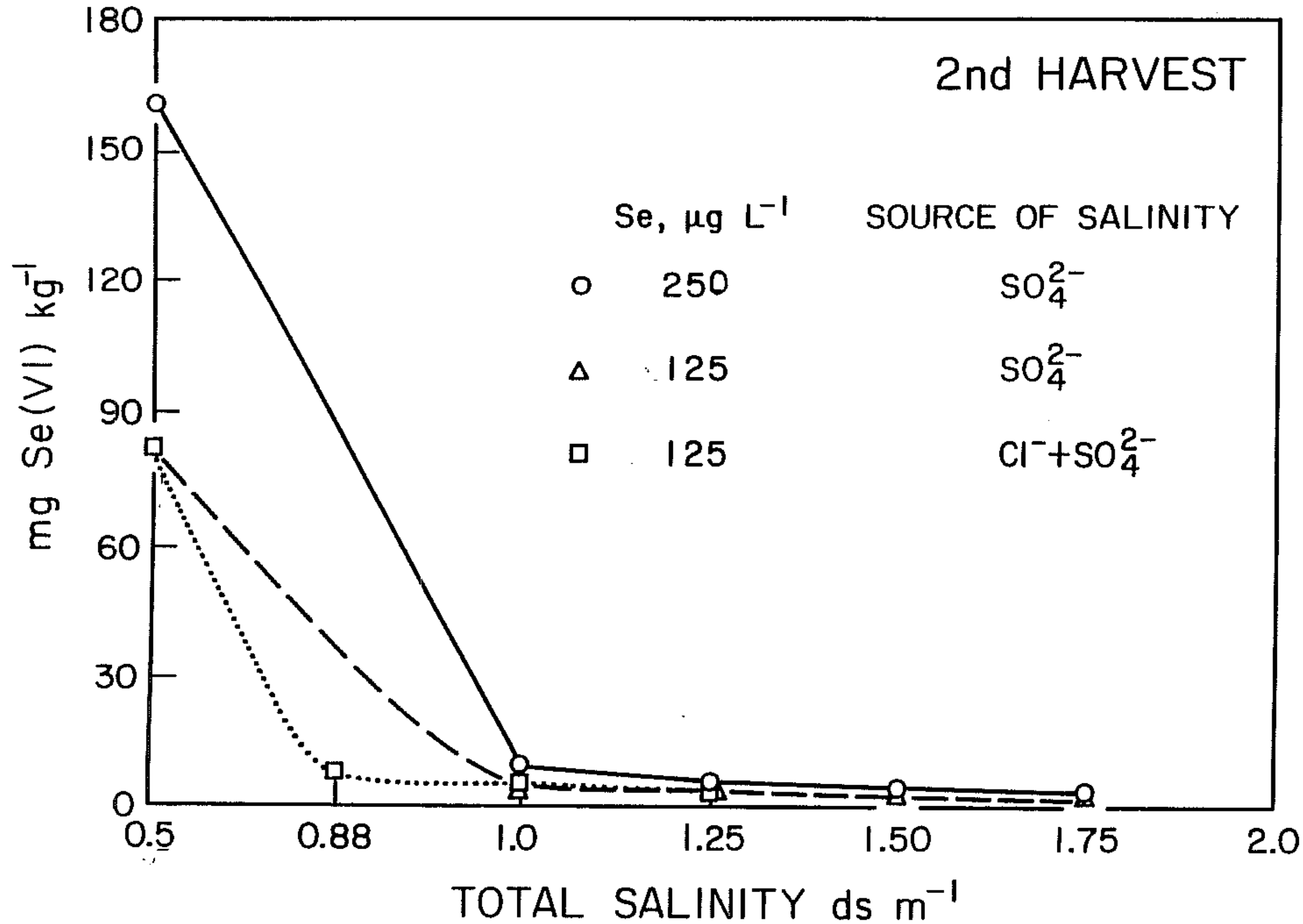


Fig. 3. EFFECTS OF SO_4 AND Cl ON Se UPTAKE BY ALFALFA

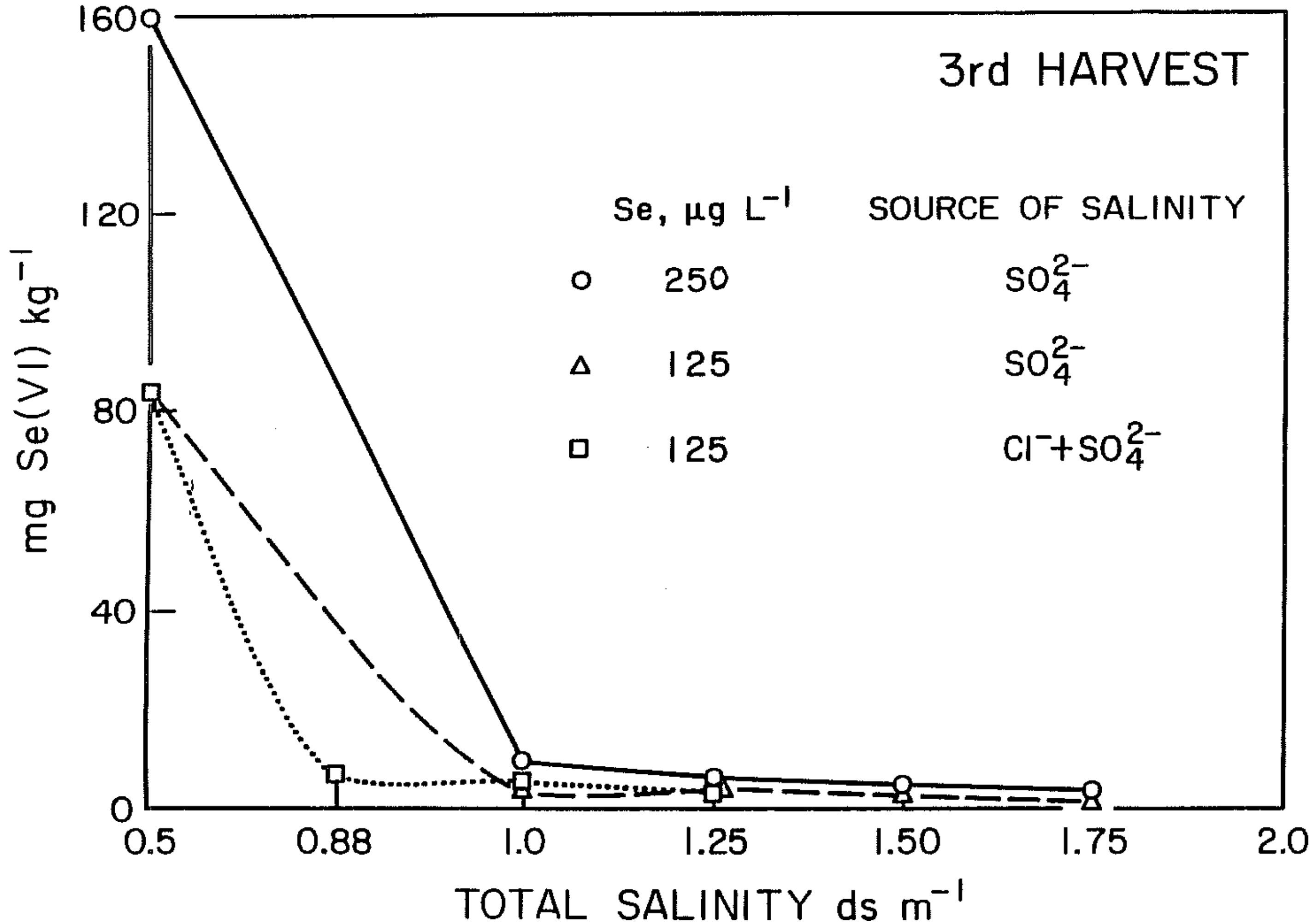
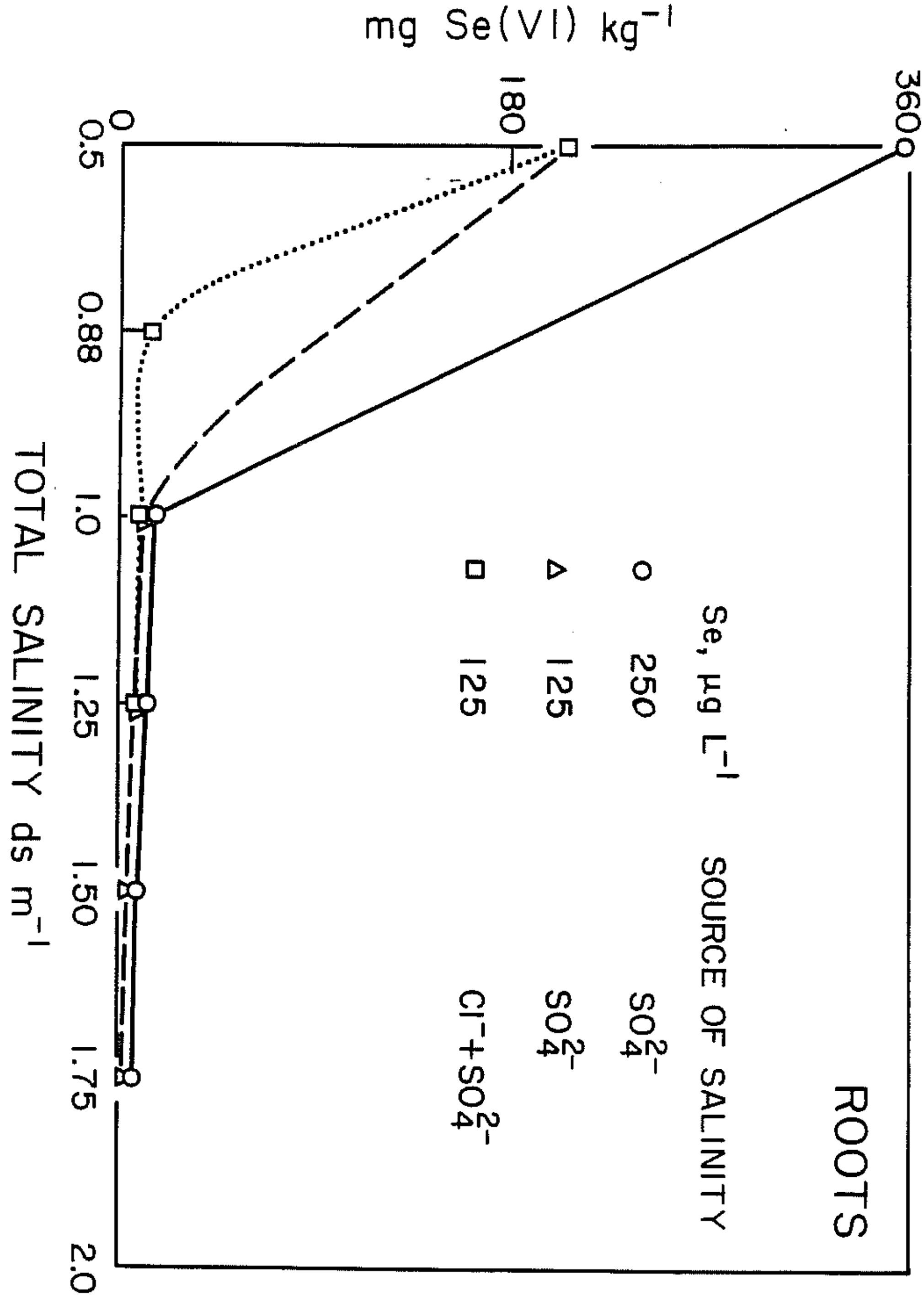


Fig. 4. EFFECTS OF SO_4 AND Cl ON Se UPTAKE BY ALFALFA



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